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What chickens would tell you about the evolution of antigen processing and presentation

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Abstract: Outside of mammals, antigen processing and presentation have only been investigated in chickens. The chicken MHC is organized differently than mammals, allowing the co-evolution of polymorphic genes, with each MHC haplotype having a set of TAP1, TAP2 and tapasin alleles directed to high expression of a single classical class I molecule. However, the class I alleles vary in the size of peptide-binding repertoire, along with a suite of other properties. The salient features of the chicken MHC are found in many non-mammalian vertebrates, and are likely to have been set at the origin of the adaptive immune system of jawed vertebrates, with unrelated genes co-evolving to set up the original pathways. Half a billion years later, various features of presentation and resistance to disease still reflect this ancestral arrangement.

Response of Jim Kaufman to Editors'comments about "What chickens would tell you about the evolution of antigen processing and presentation" 30Dec14

This a thoughtful and fascinating review of the MHC class I system in birds (or chickens, as the case may be).

I happy to hear that this ok. I would have loved to discuss all non-mammalian vertebrates at length, but I was forced to focus on the chicken, since (as mentioned in the first paragraph) all other non-mammalian vertebrates have only been examined at the level of gene sequence (except for X. laevis class I molecules which were run on SDS gels after immunoprecipitation).

Just a few comments for the authors to consider:

End of page 1. Last paragraph may be oversimplified account of what is know about mammals, since rats are more chicken like, in having fewer class I allomorphs and more allelic variation in TAP.

Quite right, and I thought that I would cover that exception by saying "humans and other typical mammals", so I could discuss the details of the fascinating rat MHC at a later place. However, I have now introduced a final sentence in that first section to explicitly make the point at the start of the piece ("As discussed in detail below, there is at least one atypical placental mammal; some of the features found in chickens were first discovered in rats.").

Authors might comment/speculate about the functional significance of class I expression on chicken erythrocytes and possible deleterious effects of producing anti-self class I antibodies (absence of which is related to egg vs. placenta?)

This is an interesting issue, but from my perspective somewhat outside of the scope of this paper, which is already too long. However, a number of points are of interest here, including the facts that: mammals alone of the vertebrates have enucleated mature erythrocytes, mammalian erythroblasts bear class I (and class II) molecules (Robinson et al 1981 Nature 289: 68), the mature erythrocytes of rats (Begovich et al 1983 Immunogenetics 18: 45; Misra et al 1983 J Immunol 10: 379) and rainbow trout (Dijkstra et al 2003 Fish Shellfish Immunol 14: 1) bear class I molecules at a low level, the mature erythrocytes of chicken (Crone et al 1985 Immunogenetics 21: 181) and the later developmental waves of frog X. laevis *bear significant numbers of class I molecules (Flajnik and Du Pasquier 1988 Devel Biol 128: 198), and later developmental waves of the erythrocytes of the salamander axolotl bear class II molecules (Volk et al 1998 Immunogenetics 47: 399). In chickens and fish, the presence of class I molecules has been tested for allogeneic responses (eg, Sarder et al 2003 Immunogenetics 55: 315). The presence of class I on rat erythrocytes is not supportive of the idea that development in eggs versus placenta is allied to these phenomena, but it doesn't rule it out either. My own guess is that the expression of class I (and class II) molecules on mature erythrocytes is left over from earlier developmental stages and is otherwise without normal functional significance, but to my knowledge this has not been tested in any system.*

Highlights for Kaufman

"What chickens would tell you about evolution of antigen processing and presentation"

There is a dominantly-expressed class I molecule for each MHC haplotype in chickens

Chicken TAP and tapasin genes are polymorphic to support one class I allele/gene

Genomic organization of chicken MHC leads to co-evolution of class I system components

The salient features of chicken MHC are shared with many non-mammalian vertebrates

Antigen processing, presentation and recognition genes co-evolved in the primordial MHC

Title: What chickens would tell you about the evolution of antigen processing and presentation

Short title: Evolution of antigen processing and presentation

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Abstract:

Outside of mammals, antigen processing and presentation have only been investigated in chickens. The chicken MHC is organized differently than mammals, allowing the co-evolution of polymorphic genes, with each MHC haplotype having a set of TAP1, TAP2 and tapasin alleles directed to high expression of a single classical class I molecule. However, the class I alleles vary in the size of peptide-binding repertoire, along with a suite of other properties. The salient features of the chicken MHC are found in many non-mammalian vertebrates, and are likely to have been set at the origin of the adaptive immune system of jawed vertebrates, with unrelated genes co-evolving to set up the original pathways. Half a billion years later, various features of presentation and resistance to disease still reflect this ancestral arrangement.

Introduction: most known components for antigen processing of class I molecules are present in chickens

Antigen presentation by classical molecules of the major histocompatibility complex (MHC) is supported by a complex set of pathways with many components [1]. Virtually all our understanding derives from a few mammals important for biomedical research: humans, mice and rats. To what extent are the antigen processing and presentation pathways conserved, both for classical MHC molecules in other organisms and for antigen presentation by other kinds of molecules? Outside of mammals, most of our knowledge about antigen presentation comes from chickens, for which there has been decades of research to support the economically-important poultry industry [2,3]. In this article, I will assume general knowledge about antigen processing and peptide loading, and focus on the differences, interpretations and ramifications of our current knowledge about the chicken class I system.

At our present level of understanding, all the expected genes encoding molecules involved in classical class I processing and presentation are present in chickens, except for inducible proteasome components, and are expected to function together in a similar way to mammals. In a compact and relatively simple MHC (a "minimal essential MHC" or "core MHC"), two class I genes (BF1 and BF2) flank transporter associated with antigen processing (TAP) 1 and TAP2 genes, with the single tapasin gene nearby [4-6]. Located outside the MHC are the genes for β_2-microglobulin (β_2m), endoplasmic reticulum amino protease (ERAP), TAP binding protein-related (TAPBPR), endoplasmic reticulum protein of 57 kDa (ERp57), calreticulin (although incomplete in the current assembly) and calnexin (Table 1). The promoters contain many of the expected transcription factor binding sites, including type I interferon element (IRES) for class I, β_2m, and TAP genes [6-11]. At least some of these components assemble into a peptide-loading complex (PLC), since class I heavy chains with β_2m co-immunopreciptate with TAP1 and TAP2 proteins [12].

It was a surprise not to find interferon ɣ-inducible proteasome component (PSMB8, β8 or LMP7, PSMB9, β9 or LMP2) genes in the sequence of the chicken MHC (the BF-BL region), and subsequent sequencing of nearby regions has failed to identify such genes [5,6]. The chicken whole genome shotgun (WGS) sequence also failed to identify those genes or PSMB10 (β10 or MECL1), thymus-specific β5t (PSMB11) and proteasome activator complex genes (PSME1 or PA28α, PSME2 or PA28β). A caveat is that the chicken genome is far from complete, but these genes were also not found in other avian genomes [13,14]. Two-dimensional gel analysis followed by mass spectrometry of immune tissues (including spleen and thymus) compared to non-immune tissues also failed to find candidate proteins, and in vitro assays failed to find differences in proteolytic specificities [14]. The chicken BF2*0401 molecule has glutamic acid as a C-terminal anchor residue, which would not be expected from a proteasome with inducible components [16]. Given that these genes have not been identified in any avian genome [15], it seems likely that the interferon-inducible as well as the thymus-specific proteasome components are simply absent. However, proteasome genes are found in the MHC of many other vertebrates, with an expanded and/or polymorphic multigene family in various reptiles, *Xenopus* frogs, bony fish and sharks [17-19].

Although nearly all the components of class I system are present in chickens and expected to function together in a similar way to mammals, overall the system works in a profoundly different way, as detailed below. In humans and other typical placental mammals, a multigene family of polymorphic class I molecules choose peptides from a large pool of peptides created by the proteasome, translocated by the TAP peptide transporter and edited by tapasin [1] (along with TAPBPR [20]). The inducible proteasome components, TAP chains and tapasin have few if any alleles with little sequence diversity and no demonstrated functional polymorphism. In contrast, only one of the two chicken classical class I genes is well-expressed, at the level of RNA, protein and antigenic

peptide. Moreover, TAP1, TAP2 and tapasin show high allelic polymorphism with moderate sequence diversity. Except for presumed recombinant(s), every MHC haplotype has a unique set of TAP1, TAP2 and tapasin alleles that are postulated to work together with the class I molecules in some (as yet incompletely understood) optimal way [4,5]. As discussed in detail below, there is at least one atypical placental mammal; some of the features found in chickens were first discovered in rats.

A dominantly-expressed class I molecule in chickens, but with a range of properties

Examination of cell surface expression level and peptides bound to chicken class I molecules revealed two kinds of MHC haplotypes (or a range between two extremes) that vary in a suite of properties [4,16,21, Chappell et al and Tregaskes et al, unpublished]. At one end are class I molecules with high cell surface expression, fastidious peptide binding and high thermal stability, and at the other end are molecules with lower cell surface expression, promiscuous peptide binding and lower thermal stability. The fastidious molecules have stringent peptide motifs, many with three anchor positions with only one or two amino acids predominating at each position [4,16]. The promiscuous molecules bind an astonishing variety of peptides. For instance, the BF2*2101 molecule remodels the binding site to allow peptides with completely dissimilar sequences to bind [21]. The BF2*0201 and BF2*1401 molecules have wide shallow pockets which allow a range of hydrophobic amino acids in the anchor positions [Chappell et al, unpublished]. This hierarchy correlates with susceptibility to Marek's disease caused by an oncogenic herpes virus, with the lower-expressing promiscuous molecules correlating with resistance. We propose that resistance correlates with breadth of peptide presentation and T cell recognition, suggesting that the phenomena of dominant peptides and CD8 T cell clonal narrowing may not be features of the responses involving promiscuous chicken class I molecules. We also propose that the expression level polymorphism evolved to optimize T cell responses in the periphery after negative selection in the thymus [Chappell et al, unpublished].

Compared to chickens, the expression of multiple class I loci in mammals averages these properties. However, several recent reports suggest that the same phenomena are present in humans. The number of self-peptides predicted to bind HLA-B57:01, B27:05, B07:02 and B35:01 varied over a five-fold range and correlated inversely with progression from HIV infection to AIDS [22]. Binding studies with peptide libraries showed that human HLA-A and HLA-B molecules vary over more than an order of magnitude in the variety of peptides bound [23]. Moreover, cell surface expression level among HLA-C alleles varies, correlating with progression to AIDS [24,25]. Most recently, semi-quantitative flow cytometry shows that expression of HLA-B57:01, B27:05, B07:02 and B35:01 on the surface of ex vivo lymphocytes and monocytes varies inversely with peptide repertoire [Chappell et al, unpublished]. The same correlation in humans and chickens suggests that these properties are fundamental features of classical class I molecules, although it appears that range of peptide repertoire is wider among chicken molecules.

Some studies show that the peptide motif of the dominantly-expressed class I molecule (BF2) can explain the strong associations of the chicken MHC with resistance and susceptibility to infectious disease [16,21,26], but the presence of a minor class I (BF1) gene is a mystery. In all haplotypes examined, BF1 RNA levels are much less than BF2, due to substitutions and/or deletions in the promoter, splicing defects or even disruption within the gene yielding a pseudogene [9,10]. The BF1 gene products have not been detected in MHC haplotypes with high expressing fastidious BF2 molecules, but in the B2 haplotype with a lower expressing promiscuous BF2 molecule, a few peptides eluted from total class I molecules fit the expected motif for the BF1 molecule and bind to BF1*0201 but not BF2*0201 [Chappell et al, unpublished]. The minor BF1*0201 molecule is fastidious, so it might be both a higher expressing and more stable molecule than the minor class I molecules of B4, B12, B19, and B21 haplotypes, which appear promiscuous based on their sequences

[16,21]. So, the minor BF1 molecules can be on the surface of cells in at least one haplotype, and therefore may be recognized by CD8 T lymphocytes. One possibility is that BF1 expression is higher and more important outside of the systemic immune system, for instance in the intestine (as hinted by RNA expression in the caecal tonsil, [9]). Another proposal [27] is that BF1 is recognised by natural killer (NK) cells in a similar way as HLA-C, considered by many to have specialized as an NK ligand. The NK receptor BNK (a homolog of NKR-P1, [5]) does not recognize class I molecules, as assessed by transfection [28]. However, a large family of chicken immunoglobulin-like receptor genes (CHIRs, chromosome 31) includes putative NK receptors homologous to human KIRs [29].

Co-evolution of interacting polymorphic genes leads to presentation by chicken class I molecules

In contrast to typical mammals, chicken TAP genes have high allelic polymorphism, moderate sequence diversity and profound functional diversity, with the translocation specificity of the TAP alleles mirroring the peptide-binding specificity of the dominantly-expressed class I molecules [11,12]. We have proposed that there is co-evolution (in the sense introduced by Germain, [30]) between the TAP and class I molecule (Fig. 1), in which optimal combinations of alleles can stay together due to the relative lack of recombination between the TAP and class I genes, in part because these genes are only tens of nucleotides apart. Humans and other typical mammals have reasonable levels of recombination between the class I genes in the class I region and the TAP genes in the class II region, so optimal combinations cannot stay together and thus the TAP genes become monomorphic, pumping a wide variety of peptides for all possible alleles and loci. However, rats have ended up with class I genes in the extended class II region, relatively close to the TAP genes. This proximity has allowed co-evolution but not as intense as in chickens, with two functional alleles of rat TAP2 that each specifies a range of amino acids at the C-terminus of the peptide, generally the same as the linked class I molecules [31,32]. Diversity is scattered throughout both chicken TAP sequences, but some polymorphic residues are found in the same positions as functionally-important polymorphisms in rat TAP2 and in sequence stretches known to contact peptides in human TAPs [11,12].

For chicken MHC haplotypes in which the peptides eluted from class I molecules indicate fastidious motifs, the TAP molecules specify as many as three positions (predominantly), each with one or two amino acids with similar chemical properties [12]. In contrast, *in vitro* assays show that the class I molecules can bind a much wider variety of peptides than are actually found at the cell surface [33, Tregaskes et al, unpublished]. Thus, the TAP translocation specificities in these haplotypes are even more stringent than the binding specificities of the class I molecule, controlling the peptides that will be presented. A consequence of this arrangement may be significant changes in the peptides presented by a class I molecule in heterozygotes compared to homozygotes, extending current concepts of heterozygote advantage.

For chicken MHC haplotypes with lower expressing promiscuous BF2 molecules, the situation appears reversed. For the B21 haplotype, the TAP molecules transport a wider variety of peptides than the dominantly-expressed class I molecule will bind [Tregaskes et al, unpublished]. However, the translocation specificity appears nowhere near as wide as for human TAPs, with the chicken B21 TAPs failing to pump peptides from other chicken haplotypes such as B4 and B15 [12]. It is not clear how this fits with a recent report that B4 and B21 chicken TAP alleles can supply sufficient peptides for unknown human class I molecules to be expressed at the cell surface [34]. However, it seems clear that minor chicken class I molecules may receive peptides in cells with TAPs from haplotypes with promiscuous class I molecules.

The single tapasin gene in chicken has as many alleles and as much sequence diversity as the TAP genes, although the variation is concentrated in a few positions, notably a loop next to the membrane in the membrane-proximal immunoglobulin-like domain [35]. *In vitro* assays involving chicken tapasin and class I molecules held together by jun-fos coiled-coils show that tapasin has effects on peptide binding of BF2 molecules, although some BF2 alleles benefit more than others. Examining the BF2 molecules by molecular dynamic simulations and statistical coupling analysis, a chain of connected residues transferring conformational information from the peptide-binding domains to the α3 domain was identified [36], tying together the both observations in mammalian class I that residues in both the α2 and α3 domains (like T134 and D220) are important in tapasin action [37,38], and the location of polymorphic residues in chicken tapasin.

Pulse-chase experiments using chicken cells transfected with wildtype and mutant class I molecules matched or mismatched with the tapasin allele showed differences in class I maturation. However, no big differences were found in the activity of different tapasin alleles *in vitro* [35]. It is not clear yet whether the lack of functional polymorphism is true for tapasin in the context of the PLC, or due to some limitation of the *in vitro* assays. For example, it may be important for chicken tapasin to be associated with ERp57, despite lacking the canonical cysteine that in mammalian tapasin forms a disulfide bond with ERp57 [39,40]. Another interesting feature of chickens is the lack of the N-terminal tapasin-binding loops in TAP1, so that the avian PLC would bind only one tapasin-class I complex at a time, compared to mammals that bind two [11,12]. This lack of TAP1 loops (but with the presence of a tapasin gene, except perhaps in ducks [41]) has been found in all avian species examined thus far, but not in any other vertebrate.

Most features of the antigen presentation genes of chickens are found in other non-mammalian vertebrates

Although the evidence is fragmentary, many of the non-mammalian vertebrates examined have the salient features discovered in chickens [42,43]. Many galliform birds have the same overall MHC structure as the chicken, and even the quail (long exemplified as a counter example [44]) has been found to have a single dominantly-expressed classical class I molecule [45]. Ducks have one well-expressed class I gene (out of five) that are located next to polymorphic TAP1 and TAP2 genes [46]. There are conflicting reports about the organization of the MHC of the passerine bird zebrafinch, but there is a single classical class I gene that is likely to be next to the TAP genes [47,48]. The MHC of *Xenopus* frogs is bigger and more complex but organized like the chicken, with polymorphic TAP genes and proteasome components near the single classical class I gene [49]. Although the MHC of bony fish is generally fragmented, class I system genes are usually together in one region; for instance, the Atlantic salmon has a single classical class I gene with a polymorphic TAP2 gene nearby [19]. In the nurse shark *Ginglymostoma cirratum*, the single classical class I gene is located close to the TAP and proteasome genes [50]. Of three marsupials examined, the MHC of the American opossum is organized like the chicken [51].

However, there are exceptions. The Tammar wallaby MHC contains the TAP genes, but all of the many classical class I genes are found at the telomeres of other chromosomes [52,53]. The Tasmanian devil MHC is incompletely assembled but expresses two or three classical class I genes in each haplotype [56]. Salamanders such as the axolotl and *Gadus* fish such as Atlantic cod express numerous class I genes which are considered to be classical class I molecules, although nothing is known about their real functions. There remains the possibility that some of these class I genes have new functions (for instance, replacing the missing class II system in the Atlantic cod) [54-57].

The likeliest scenario is that the organization first described in chickens is the ancestral organization. In this view [12,43,58], the MHC of typical placental mammals is derived by an inversion that brought the class III region into the centre of the MHC, and swept the class I gene(s) to the outside, but with the breakpoint such that the antigen processing genes (such as the TAP, tapasin and inducible proteasome genes) were left behind and by default ended up in the class II region. Once the class III region separated the genetic components of the class I system, advantageous combinations of alleles were separated by recombination and the co-evolutionary relationships were lost (Fig. 1). The result in placental mammals (and expected for the Tammar wallaby) was that the antigen processing genes became monomorphic, able to generate, pump and load peptides for all class I alleles switched around by recombination, eventually allowing a class I multigene family.

Conclusions: how could a co-evolving system originate and continue to evolve?

The concept of co-evolution between nearby genes can neatly explain some phenomena of the chicken class I system, but does it have selective advantages or is it an accident of history? One potential selective disadvantage would be the necessity of compensatory mutations in one gene to match any changes in another gene, slowing down the rate of MHC evolution. As a possible explanation, perhaps promiscuous TAP and class I genes are pre-adapted (or exapted, to use the more specialised term) for switching specificities.

Another question is whether the principles discovered for the chicken class I system hold for other systems of antigen presentation. To take a closely-related example, there is a single nearly monomorphic DR-like class II A gene located over 5 cM away from the chicken MHC, and two polymorphic class II B genes located in the MHC, one of which is expressed far more than the other [59,60]. Nearby are the polymorphic DM genes, with a single DMA gene and two DMB genes, only one of which is well-expressed [5,6,61]. Superficially, the chicken class II system has many parallels with the chicken class I system. Are these similarities based on the same principles, and are these features found in other non-mammalian vertebrates? Further afield, the discoveries that elephant sharks lost the CD4 gene (and potentially possess only the Th1 subset) and that Atlantic cod and other *Gadus* fish lost the class II system altogether deserve more investigation [56,62]. Also, more detailed examination of other antigen presentation systems (CD1 with lipids, MR1 and butyrophilin with metabolites) should prove extremely interesting [63-66].

Finally, if the features of the chicken classical class I system are ancestral, then why should this be so? One explanation might be that at the origin of antigen presentation, genes with disparate structures and functions had to co-evolve to create the pathway of antigen presentation; the most efficient way would be for these genes to be closely-linked so that advantageous combinations could stay together. This scenario [12,43,58] is much like the model proposed for the evolution of metabolic pathways, and would explain the presence of antigen processing and loading genes in the MHC as originally necessary for development of antigen processing pathways, and the subsequent loss of such genes from the MHC by genome-wide duplication followed by selective silencing (for example, the inducible proteasome component MECL1 in humans), or by translocation. A further speculation [49,67], based on the presence of NK cell receptor B-NK in the chicken MHC and later the presence of a T cell receptor-like gene at the outskirts of the *Xenopus* MHC, is that not only ligands but receptors had to be present to co-evolve. In this view, the concept of co-evolution between nearby genes not frequently separated by recombination may be an important theme in the evolution of the whole adaptive immune system.

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Figure Legend.

Figure 1. Polymorphic interacting molecules must be encoded by closely-linked genes for optimal combinations to remain together. Red genes and molecules share peptide specificity, and blue genes and molecules share a different specificity. Left, TAP and class I genes located close together as in the chicken (and many non-mammalian vertebrates), so that there is little recombination to split optimal combinations (blue TAP molecules provide the appropriate peptides for blue class I molecules). Right, TAP and class I genes located far apart as in a typical mammal, so that recombination followed by reassortment leads to inappropriate combinations (blue TAP molecules provide the inappropriate peptides for red class I molecules).

References.

1.** Blum JS, Wearsch PA, Cresswell P: Pathways of antigen processing. Annu Rev Immunol 2013, 31: 443-473.

**A clear view of current thoughts about antigen processing as discovered in humans and mice

2.** Kaufman J: Antigen processing and presentation: evolution from a bird's eye view. Molec Immunol 2013, 55:159-61.

**A quick summary of differences between the birds and mammals in terms of structure, function and evolution

3. Schat AK, Kaspers B, Kaiser P (eds): Avian Immunology second edition. Academic Press; 2014.

4. Kaufman J, Völk H, Wallny H-J: A "minimal essential Mhc" and an "unrecognized Mhc": two extremes in selection for polymorphism. Immunol Rev 1995, 143: 63-88.

5. Kaufman J, Milne S, Göbel TW, Walker BA, Jacob JP, Auffray C, Zoorob R, Beck S: The chicken B locus is a minimal essential major histocompatibility complex. Nature 1999, 401: 923-925.

6. Hosomichi K, Miller MM, Goto RM, Wang Y, Suzuki S, Kulski JK, Nishibori M, Inoko H, Hanzawa K, Shiina T: Contribution of mutation, recombination, and gene conversion to chicken MHC-B haplotype diversity. J Immunol 2008, 181: 3393-3399.

7. Kroemer G, Zoorob R, Auffray C: Structure and expression of a chicken MHC class I gene. Immunogenetics 1990, 31: 405-409.

8. Riegert P, Andersen R, Bumstead N, Döhring C, Dominguez-Steglich M, Engberg J, Salomonsen J, Schmid M, Schwager J, Skjødt K, Kaufman J: The chicken beta2-microglobulin gene is located on a non-major histocompatibility complex microchromosome: a small, G+C-rich gene with X and Y boxes in the promoter. Proc Natl Acad Sci U S A 1996, 93: 1243-1248.

9. Shaw I, Powell TJ, Marston DA, Baker K, van Hateren A, Riegert P, Wiles MV, Milne S, Beck S, Kaufman J: Different evolutionary histories of the two classical class I genes BF1 and BF2 illustrate drift and selection within the stable MHC haplotypes of chickens. J Immunol 2007, 178: 5744-5752.

10. O'Neill AM, Livant EJ, Ewald SJ: The chicken BF1 (classical MHC class I) gene shows evidence of selection for diversity in expression and in promoter and signal peptide regions. Immunogenetics 2009, 61: 289-302.

11. Walker BA, van Hateren A, Milne S, Beck S, Kaufman J: Chicken TAP genes differ from their human orthologues in locus organisation, size, sequence features and polymorphism. Immunogenetics 2005, 57: 232-247.

12. Walker BA, Hunt LG, Sowa AK, Skjødt K, Göbel TW, Lehner PJ, Kaufman J: The dominantly expressed class I molecule of the chicken MHC is explained by coevolution with the polymorphic peptide transporter (TAP) genes. Proc Natl Acad Sci U S A 2011, 108: 8396-8401.

13. International Chicken Genome Sequencing Consortium: Sequencing and comparative analysis of the chicken genome. Nature 2004, 432: 695-716.

14. Sutoh Y, Kondo M, Ohta Y, Ota T, Tomaru U, Flajnik MF, Kasahara M: Comparative genomic analysis of the proteasome β5t subunit gene: implications for the origin and evolution of thymoproteasomes. Immunogenetics 2012, 64: 49-58.

15.* Erath S, Groettrup M: No evidence for immunoproteasomes in chicken lymphoid organs and activated lymphocytes. Immunogenetics 2014, Epub ahead of print, PubMed PMID: 25403261.

*Evidence at the biochemical level for the lack of inducible proteasome components in chickens

16. Wallny HJ, Avila D, Hunt LG, Powell TJ, Riegert P, Salomonsen J, Skjødt K, Vainio O, Vilbois F, Wiles MV, Kaufman J: Peptide motifs of the single dominantly expressed class I molecule explain the striking MHC-determined response to Rous sarcoma virus in chickens. Proc Natl Acad Sci U S A 2006, 103: 1434-1439.

17. Nonaka M, Yamada-Namikawa C, Flajnik MF, Du Pasquier L: Trans-species polymorphism of the major histocompatibility complex-encoded proteasome subunit LMP7 in an amphibian genus, Xenopus. Immunogenetics 2000, 51: 186-192.

18. Tsukamoto K, Miura F, Fujito NT, Yoshizaki G, Nonaka M: Long-lived dichotomous lineages of the proteasome subunit beta type 8 (PSMB8) gene surviving more than 500 million years as alleles or paralogs. Mol Biol Evol 2012, 29: 3071-3079.

19. Lukacs MF, Harstad H, Grimholt U, Beetz-Sargent M, Cooper GA, Reid L, Bakke HG, Phillips RB, Miller KM, Davidson WS, Koop BF: Genomic organization of duplicated major histocompatibility complex class I regions in Atlantic salmon (Salmo salar). BMC Genomics 2007, 8: 251. PubMed PMID: 17651474; PubMed Central PMCID: PMC1971071.

20. Boyle LH, Hermann C, Boname JM, Porter KM, Patel PA, Burr ML, Duncan LM, Harbour ME, Rhodes DA, Skjødt K, Lehner PJ, Trowsdale J: Tapasin-related protein TAPBPR is an additional component of the MHC class I presentation pathway. Proc Natl Acad Sci U S A 2013, 110: 3465-3470.

21. Koch M, Camp S, Collen T, Avila D, Salomonsen J, Wallny HJ, van Hateren A, Hunt L, Jacob JP, Johnston F, Marston DA, et al: Structures of an MHC class I molecule from B21 chickens illustrate promiscuous peptide binding. Immunity 2007, 27: 885-899

22. Kosmrlj A, Read EL, Qi Y, Allen TM, Altfeld M, Deeks SG, Pereyra F, Carrington M, Walker BD, Chakraborty AK: Effects of thymic selection of the T-cell repertoire on HLA class I-associated control of HIV infection. Nature 2010, 465: 350-354.

23.* Paul S, Weiskopf D, Angelo MA, Sidney J, Peters B, Sette A: HLA class I alleles are associated with peptide-binding repertoires of different size, affinity, and immunogenicity. J Immunol 2013, 191: 5831-5839.

*Evidence at the biochemical level for differences in peptide-repertoires of human class I molecules

24. Thomas R, Apps R, Qi Y, Gao X, Male V, O'hUigin C, O'Connor G, Ge D, Fellay J, Martin JN, Margolick J, et al: HLA-C cell surface expression and control of HIV/AIDS correlate with a variant upstream of HLA-C. Nat Genet 2009, 41: 1290-1294.

25.* Apps R, Qi Y, Carlson JM, Chen H, Gao X, Thomas R, Yuki Y, Del Prete GQ, Goulder P, Brumme ZL, Brumme CJ, et al: Influence of HLA-C expression level on HIV control. Science 2013, 340: 87-91.

*Evidence for polymorphism of class I expression at the cell surface correlating with disease resistance

26. Butter C, Staines K, van Hateren A, Davison TF, Kaufman J: The peptide motif of the single dominantly expressed class I molecule of the chicken MHC can explain the response to a molecular defined vaccine of infectious bursal disease virus (IBDV). Immunogenetics 2013, 65: 609-618.

27. Ewald SJ, Livant EJ: Distinctive polymorphism of chicken B-FI (major histocompatibility complex class I) molecules. Poult Sci 2004, 83: 600-605.

28. Viertlboeck BC, Wortmann A, Schmitt R, Plachý J, Göbel TW: Chicken C-type lectin-like receptor B-NK, expressed on NK and T cell subsets, binds to a ligand on activated splenocytes. Mol Immunol 2008, 45: 1398-1404.

29. Straub C, Neulen ML, Sperling B, Windau K, Zechmann M, Jansen CA, Viertlboeck BC, Göbel TW: Chicken NK cell receptors. Dev Comp Immunol 2013, 41: 324-333.

30. Germain RN, Bentley DM, Quill H: Influence of allelic polymorphism on the assembly and surface expression of class II MHC (Ia) molecules. Cell 1985, 43: 233–242.

31. Powis SJ, Young LL, Joly E, Barker PJ, Richardson L, Brandt RP, Melief CJ, Howard JC, Butcher GW: The rat cim effect: TAP allele-dependent changes in a class I MHC anchor motif and evidence against C-terminal trimming of peptides in the ER. Immunity 1996, 4: 159-165.

32. Joly E, Le Rolle AF, González AL, Mehling B, Stevens J, Coadwell WJ, Hünig T, Howard JC, Butcher GW. Co-evolution of rat TAP transporters and MHC class I RT1-A molecules. Curr Biol 1998, 8: 169-172.

33.* Zhang J, Chen Y, Qi J, Gao F, Liu Y, Liu J, Zhou X, Kaufman J, Xia C, Gao GF: Narrow groove and restricted anchors of MHC class I molecule BF2*0401 plus peptide transporter restriction can explain disease susceptibility of B4 chickens. J Immunol 2012, 189: 4478-4487.

*Evidence for restriction of class I presentation by TAP translocation specificity

34.* Hinz A, Jedamzick J, Herbring V, Fischbach H, Hartmann J, Parcej D, Koch J, Tampé R: Assembly and function of the major histocompatibility complex (MHC) I peptide-loading complex are conserved across higher vertebrates. J Biol Chem 2014, 289: 33109-33117.

*Surprising ability of a chicken TAP transporter with very specific translocation motifs to support cell surface expression of undefined human class I molecules

35.* van Hateren A, Carter R, Bailey A, Kontouli N, Williams AP, Kaufman J, Elliott T: A mechanistic basis for the co-evolution of chicken tapasin and major histocompatibility complex class I (MHC I) proteins. J Biol Chem 2013, 288: 32797-32808.

*The first examination at the biochemical level of the effects of chicken tapasin alleles on class I peptide editing

36.* Bailey A, van Hateren A, Elliott T, Werner JM. Two polymorphisms facilitate differences in plasticity between two chicken major histocompatibility complex class I proteins. PLoS One 2014 (doi:10.1371/journal.pone.0089657), 9: e89657.

*Evidence for information transfer between the peptide-binding domain and the α3 domain during peptide-editing

37. Lewis JW, Neisig A, Neefjes J, Elliott T: Point mutations in the alpha 2 domain of HLA-A2.1 define a functionally relevant interaction with TAP. Curr Biol 1996, 6: 873-883.

38. Turnquist HR, Vargas SE, Schenk EL, McIlhaney MM, Reber AJ, Solheim JC: The interface between tapasin and MHC class I: identification of amino acid residues in both proteins that influence their interaction. Immunol Res 2002, 25: 261-269.

39. Frangoulis B, Park I, Guillemot F, Séverac V, Auffray C, Zoorob R: Identification of the Tapasin gene in the chicken major histocompatibility complex. Immunogenetics 1999, 49: 328-337.

40. Dong G, Wearsch PA, Peaper DR, Cresswell P, Reinisch KM: Insights into MHC class I peptide loading from the structure of the tapasin-ERp57 thiol oxidoreductase heterodimer. Immunity 2009; 30: 21-32.

41. Magor KE, Miranzo Navarro D, Barber MR, Petkau K, Fleming-Canepa X, Blyth GA, Blaine AH: Defense genes missing from the flight division. Dev Comp Immunol 2013, 41: 377-388.

42. Kaufman J: Co-evolving genes in MHC haplotypes: the "rule" for nonmammalian vertebrates? Immunogenetics 1999, 50: 228-236.

43.** Kaufman J: The Avian MHC. In Avian Immunology, second edition. Edited by Schat KA, Kaiser P, Kaspers B. Elsevier, Ltd; 2014: 149-167.

**Comprehensive description of the chicken MHC and models arising for evolution of the adaptive immune system

44. Shiina T, Shimizu S, Hosomichi K, Kohara S, Watanabe S, Hanzawa K, Beck S, Kulski JK, Inoko H: Comparative genomic analysis of two avian (quail and chicken) MHC regions. J Immunol 2004, 172: 6751-6763.

45. Shiina T, Hosomichi K, Hanzawa K: Comparative genomics of the poultry major histocompatibility complex. Anim Sci J 2006, 77: 151-162.

46. Mesa CM, Thulien KJ, Moon DA, Veniamin SM, Magor KE: The dominant MHC class I gene is adjacent to the polymorphic TAP2 gene in the duck, Anas platyrhynchos. Immunogenetics 2004, 56: 192-203.

47. Balakrishnan CN, Ekblom R, Völker M, Westerdahl H, Godinez R, Kotkiewicz H, Burt DW, Graves T, Griffin DK, Warren WC, Edwards SV: Gene duplication and fragmentation in the zebra finch major histocompatibility complex. BMC Biol 2010 (doi: 10.1186/1741-7007-8-29), 8: 29.

48. Ekblom R, Stapley J, Ball AD, Birkhead T, Burke T, Slate J: Genetic mapping of the major histocompatibility complex in the zebra finch (Taeniopygia guttata). Immunogenetics 2011, 63: 523-530.

49. Ohta Y, Goetz W, Hossain MZ, Nonaka M, Flajnik MF: Ancestral organization of the MHC revealed in the amphibian Xenopus. J Immunol 2006, 176: 3674-3685.

50. Ohta Y, McKinney EC, Criscitiello MF, Flajnik MF: Proteasome, transporter associated with antigen processing, and class I genes in the nurse shark Ginglymostoma cirratum: evidence for a stable class I region and MHC haplotype lineages. J Immunol 2002, 168: 771-781.

51. Belov K, Deakin JE, Papenfuss AT, Baker ML, Melman SD, Siddle HV, Gouin N, Goode DL, Sargeant TJ, Robinson MD, Wakefield MJ, et al: Reconstructing an ancestral mammalian immune supercomplex from a marsupial major histocompatibility complex. PLoS Biol 2006, 4: e46. PubMed PMID: 16435885; PubMed Central PMCID: PMC1351924.

52. Deakin JE, Siddle HV, Cross JG, Belov K, Graves JA: Class I genes have split from the MHC in the tammar wallaby. Cytogenet Genome Res 2007, 116: 205-211.

53. Siddle HV, Deakin JE, Coggill P, Hart E, Cheng Y, Wong ES, Harrow J, Beck S, Belov K: MHC-linked and un-linked class I genes in the wallaby. BMC Genomics 2009 (doi: 10.1186/1471-2164-10-310), 10: 310.

54. Cheng Y, Stuart A, Morris K, Taylor R, Siddle H, Deakin J, Jones M, Amemiya CT, Belov K: Antigen-presenting genes and genomic copy number variations in the Tasmanian devil MHC. BMC Genomics 2012 (doi: 10.1186/1471-2164-13-87), 13: 87.

55. Sammut B, Du Pasquier L, Ducoroy P, Laurens V, Marcuz A, Tournefier A: Axolotl MHC architecture and polymorphism. Eur J Immunol 1999, 29: 2897-2907.

56. Star B, Nederbragt AJ, Jentoft S, Grimholt U, Malmstrøm M, Gregers TF, Rounge TB, Paulsen J, Solbakken MH, Sharma A, Wetten OF, et al: The genome sequence of Atlantic cod reveals a unique immune system. Nature 2011, 477: 207-210.

57. Malmstrøm M, Jentoft S, Gregers TF, Jakobsen KS: Unraveling the evolution of the Atlantic cod's (Gadus morhua L.) alternative immune strategy. PLoS One 2013, 8: e74004. PubMed PMID: 24019946; PubMed Central PMCID: PMC3760826.

58. Kaufman J: The evolutionary origins of the adaptive immune system of jawed vertebrates (Chapter 3). In The Immune Response to Infection. Edited by Kaufmann SHE, Rouse BT, Sachs DL. American Society of Microbiology Press, 2011: 41-54.

59. Salomonsen J, Marston D, Avila D, Bumstead N, Johansson B, Juul-Madsen H, Olesen GD, Riegert P, Skjødt K, Vainio O, Wiles MV, et al: The properties of the single chicken MHC classical class II alpha chain (B-LA) gene indicate an ancient origin for the DR/E-like isotype of class II molecules. Immunogenetics 2003, 55: 605-614.

60. Jacob JP, Milne S, Beck S, Kaufman J: The major and a minor class II beta-chain (B-LB) gene flank the tapasin gene in the B-F /B-L region of the chicken major histocompatibility complex. Immunogenetics 2000, 51: 138-147.

61. Chazara O, Tixier-Boichard M, Morin V, Zoorob R, Bed'hom B: Organisation and diversity of the class II DM region of the chicken MHC. Mol Immunol 2011, 48: 1263-1271.

62. Venkatesh B, Lee AP, Ravi V, Maurya AK, Lian MM, Swann JB, Ohta Y, Flajnik MF, Sutoh Y, Kasahara M, Hoon S, et al: Elephant shark genome provides unique insights into gnathostome evolution. Nature 2014, 505: 174-179.

63. De Libero G, Mori L: Novel insights into lipid antigen presentation. Trends Immunol 2012, 33: 103-111.

64. León L, Tatituri RV, Grenha R, Sun Y, Barral DC, Minnaard AJ, Bhowruth V, Veerapen N, Besra GS, Kasmar A, Peng W, et al: Saposins utilize two strategies for lipid transfer and CD1 antigen presentation. Proc Natl Acad Sci U S A 2012, 109: 4357-4364.

65. Kjer-Nielsen L, Patel O, Corbett AJ, Le Nours J, Meehan B, Liu L, Bhati M, Chen Z, Kostenko L, Reantragoon R, Williamson NA, et al: MR1 presents microbial vitamin B metabolites to MAIT cells. Nature 2012, 491: 717-723.

66. Sandstrom A, Peigné CM, Léger A, Crooks JE, Konczak F, Gesnel MC, Breathnach R, Bonneville M, Scotet E, Adams EJ: The intracellular B30.2 domain of butyrophilin 3A1 binds phosphoantigens to mediate activation of human Vγ9Vδ2 T cells. Immunity 2014, 40: 490-500.

67. Rogers SL, Göbel TW, Viertlboeck BC, Milne S, Beck S, Kaufman J: Characterization of the chicken C-type lectin-like receptors B-NK and B-lec suggests that the NK complex and the MHC share a common ancestral region. J Immunol 2005, 174: 3475-3483.

Table 1. Components of the chicken classical class I system

Component	ensemble identifier	location (chromosome, nucleotides)
Class I: BF1	ENSGALG00000000178	chromosome 16: 73,099-75,588
Class I: BF2	ENSGALG00000024372	chromosome 16: 59,616-62,099
TAP1	ENSGALG00000026269	chromosome 16: 67,623-76,124
TAP2	ENSGALG00000000181	chromosome 16: 64,031-67,067
Tapasin (TPN)	ENSGALG00000008022	chromosome 16: 97,656-101,192
β_2m	ENSGALG00000002160	chromosome 10: 827,900-828,534
TAPBPR (TAPBPL)	ENSGALG00000014428	chromosome 1: 76,360,081-76,368,699
ERAP1	ENSGALG00000014684	chromosome Z: 56,827,503-56,843,435
ERp57 (PDIA3)	ENSGALG00000008348	chromosome 10: 19,625,880-19,635,020
calreticulin (CALR)	ENSGALG00000026852	scaffold AADN03021535.1: 21-1,050
calnexin (CANX)	ENSGALG00000005945	chromosome 13: 12,858,024-12,874,898

Figure

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